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Docket No. SP02-143 (015275-060007)

Application Serial No.: 10/602,242

REMARKS

In response to the Final Office Action mailed April 18, 2007, reconsideration of the application is requested in view of the pending claims and following remarks.

Claims 1, 2, 4, 5, 9-18, and 42-61 are pending. Claims 3, 6-8 and 19-41 were previously cancelled. Claims 1, 2, 4-5, 9-15, 17, 42-44, 47, 49, and 51-61 have been amended for clarity, as discussed hereinbelow. No new matter has been introduced in this amendment.

By this amendment, independent claims 1, 42, 49, and 57 are amended by eliminating the article "an" and replacing it with the phrase "a dried" before the term array (first occurrence) and inserting the term "lipid" between the terms "biological" and "membranes" as appeared in each of the claims. In addition, claim 1 has been amended by deleting the phrase "said biological membranes are deposited directly onto said coating" and replacing it with the phrase "wherein said biological lipid membranes comprise a mixture of a host lipid and a doped lipid deposited onto the surface." Also in claim 1, the phrase "at least one biological membrane with said target compound" is replaced with the phrase "at least one of the biological lipid membranes with the toxin in the sample."

With respect to claims 42 and 49, the phrase "said biological membranes are deposited directly to said coating" is replaced with the phrase "a mixture of a host lipid and a doped lipid deposited onto the surface."

Claim 2 is amended by replacing the phrase "biological membranes" with the phrase "doped lipid." In claims 42 and 49, the phrase "biological lipid membrane" is inserted before the term "microspots" at the last line of the claims. Similarly, the phrase "biological lipid membrane" is inserted before the term "microspots" at the second line of claim 46.

To provide proper antecedent basis, the phrase "target compound" is substituted with the term "toxin" in claims 1, 12, and 13. In claim 14, the phrase "the toxin molecule or to the receptor site" is substituted with the phrase "to the toxin or to a receptor site of the toxin."

To provide clarity, the term "compound" is inserted after the term "target" in claim 43 and in claim 46 (first occurrence). To ensure proper claim dependency, the phrase "claim 42" is replaced with "claim 43" in claim 44.

In claim 57, the phrases "directly deposited to a coating on a surface of said array," and "comprising a receptor of interest" are replaced with the phrases "associated with a

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surface of a substrate, wherein" and "comprises a mixture of a host lipid and a receptor of interest and is deposited onto a coating of said surface of said substrate," respectively. In addition, the phrase "receptors in" is inserted in before the phrase "between one or more," as appeared in the last recited step of the claimed method.

The term "said" is replaced with the article "the" in claims 1, 2, 4-5, 9-12, 14-15, 17, 42, 49, and 51-61. The term "lipid" is inserted between the terms "biological" and "membranes" in claims 9 and 12.

The amendments are supported by the entire specification, particularly at p. 7, ¶ [0029], l. 1-p. 8, l. 9; p. 11, ¶ [0040], l. 1-p. 12, ¶ [0043], l. 12; p. 12, ¶ [0043], l. 1-p. 13, ll. 1-14, p. 12, ¶ [0041], l. 2, p. 8, ll. 3-7 and Figures 1-8. Accordingly, Applicants respectfully request that the Examiner enter these amendments, which clarify the subject matter of the present invention.

Applicants submit that the rejections based on lack of novelty and obviousness are also overcome in view of the amendments and arguments presented in the response. Accordingly, entry of these amendments is requested.

Claim Rejections under 35 U.S.C. §102(b)

In page 2, section 3 of the Office Action, the Examiner maintained his rejection of claims 1, 2, 4-5 and 17-18 under 35 U.S.C. § 102(b) as being anticipated by U. S. Patent No. 4,931,498 to Pidgeon (hereinafter "Pidgeon"). In particular, the Examiner, while referring to independent claim 1, asserted that Pidgeon "teaches supports having a pellicular coating formed from a polyamine, such as polyethylene (col. 6, lines 15-30) upon which biological membranes are immobilized (col. 6, lines 32-50)." Also, the Examiner contended that Pidgeon "teaches that the membranes can remove endotoxins from contaminated protein samples (col. 13, lines 1-5)." This rejection is respectfully traversed since Pidgeon fails to describe the features set forth in amended claim 1, as well as the rejected claims depending therefrom.

Independent claim 1, as amended, is directed to a method for detecting and identifying a toxin in a sample which includes (1) providing a dried array comprising a plurality of biological lipid membranes associated with a surface of a substrate, wherein the surface comprises a coating of an amine-presenting molecule wherein the biological lipid membranes comprise a mixture of a host lipid and a doped lipid deposited onto the surface;

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(2) contacting the array with a solution comprising a toxin; and (3) monitoring for binding activity of at least one of the biological lipid membranes with the toxin in the sample. Support for the claimed features can be found at least on, *e.g.*, at page 7, ¶ [0029], line 1-page 8, line 9; page 11, ¶ [0040], line 1-page 12, ¶ [0043], line 12; page 12, ¶ [0043], line 1-page 13, lines 1-14.

Pidgeon neither teaches nor suggests the method for detecting and identifying a toxin in a sample, as set forth in amended claim 1 and its dependent claims. Accordingly, Applicants respectfully submit that Pidgeon fails to anticipate claims 1, 2, 4-5 and 17-18, particularly in light of the additional remarks provided hereinbelow.

Pidgeon, as its title suggests, is directed to a covalently-immobilized membrane composition adapted for chromatographic systems, which include the following elements: (1) a particulate support material; (2) an artificial membrane structure; and (3) a means for immobilizing the membrane structure on the surface of the support material (col. 21-22 and claim 1). According to Pidgeon, "immobilization of the membrane structure can be accomplished either by covalent bonding of the component amphiphilic molecules, by hydrophobic interaction of the amphiphilic molecules with a hydrophobic surface on the particulate support structure or by a combination of hydrophobic interaction and covalent bonding." See Pidgeon at col. 4, ll. 25-32.

The "means" for immobilizing the membrane structure on the surface of the support material includes divalent functional groups covalently bonded to the surface of the support material and to the amphiphilic molecules forming the membrane structure (col. 22 and claim 2). As set forth in Pidgeon, "the preferred covalently immobilized membrane chromatographic supports in accordance with this invention can be accomplished by utilizing a novel phospholipid carboxylates derived by reaction of C₁₀-C₁₆ cyclic dicarboxylic acid anhydrides with glycerol-phosphates and lysophospholipids" (col. 3, ll. 5-10 and col. 9, ll. 10 - 16). The covalently-bound artificial membranes, as set forth in Pidgeon, are illustrated in Figures 4 and 5 and further discussed in detail in the specification text (col. 8, l. 31 to col. 11, l. 64).

According to Pidgeon, the cyclic anhydride of 15-ring member cyclic anhydride represented by Formula II (at col. 9) can be used as a source of a divalent linker in the production of immobilized membranes, wherein the linker itself is used to distance the membrane forming molecules from the support surface to provide a more "bilayer-like"

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immobilized membrane structure. (col. 11, ll. 27-36). The covalently-bound immobilized structures were described as comparable to one-half of a biological membrane bi-layer bound to the support surface.

Thus, to fully construct a full membrane bi-layer using a cyclic anhydride as a divalent linking group between the support surface and the membrane forming amphiphilic molecules, an immobilized membrane composition, as summarized by Pidgeon, at col. 11, ll. 42-58, is prepared by the following steps: "(1) react Nucleosil-300(7NH₂) with, for example a cyclic dicarboxylic anhydride of the Formula II wherein n is 14 to provide a support material having a surface of covalently bound carboxy groups some 16-18 carbon atoms (including carbon atoms in the propylamine group) from the surface of the support structure; (2) react the product supports with 1,1-carbonyldiimidazole to form the corresponding imidazolid; (3) reacting the imidazolid-bearing support material with an excess (to minimize crosslinking) of ethylenediamine so that the linker can on the surface of the Nucleosil-300(7NH₂) is represented by a group of the formula -CH₂CH₂CH₂NHCO(C₂)₁₋₂-CONHCH₂H₂CH₂; and (4) reacting the Product with. For example, the lecithin imidazolid prepared in the example below" (For additional details, see col. 15, l. 64-col. 20, l. 7).

Accordingly, as per Pidgeon, a biological membrane (*i.e.*, an artificial membrane structure) cannot be directly deposited to a coating of an amine-presenting molecule (*i.e.*, supporting material) without the inclusion of the immobilization means. As a consequence, the biological membranes employed in Pidgeon cannot be directly deposited to a coating of an amine-presenting molecule.

In contrast, the biological lipid membrane of the presently claimed invention, as set forth in amended claim 1, is made up of a mixture of a host lipid and a doped lipid, both of which are added together before being deposited onto the surface of the substrate that is coated with an amine-presenting molecule. After deposition, the biological lipid membranes are incubated under control humidity, and dried for 1 hour to "enable possible lateral redistribution of the lipid molecules in the supported membrane' before coming in contact with a target compound. See the specification at p. 12, ¶ [0043], l. 1-p. 13, ll. 1-11.

Based on the above description and contrary to Pidgeon, the biological lipid membrane of the claimed invention does not employ any covalent immobilization means or attached to any covalent functional linkers. In fact, the specification discloses that "a unique aspect of the membrane microarrays is the need to keep the probe confined to the microspot

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while maintaining the desired lateral movement of individual molecules within the microspot – properties that are contradictory and preclude covalent immobilization of the membrane.” See the specification at page 11, ¶ [0040], line 1-page 13, lines 4-7. By directly depositing the mixture of host lipid and doped lipid into the amine-presenting moiety-coated surface of the substrate array, there is no need for any functional linkers, as presently claimed.

Moreover, this direct deposition increases the surface resistance to physical desorption and improve the membrane's long-range lateral fluidity. See the specification at pages 11-12, paragraphs 40 and 42. In fact, lateral fluidity of the membrane microarrays (slides with printed microspots and doped with fluorescently labeled lipids) according to the present invention was tested and shown using fluorescence microscopy. See the specification at p. 12, ¶ [0042].

“A claim is anticipated only if each and every element as set forth in the claim is found, either expressly or inherently described, in a single prior art reference.” *Verdegaal Bros. v. Union Oil Co. of California*, 814 F.2d 628, 631, 2 USPQ2d 1051, 1053 (Fed. Cir. 1987) and MPEP 2131. In view of the above, Applicants respectfully submit that Pidgeon fails to anticipate claims 1, 2, 4-5 and 17-18.

In page 3, section 8 of the Office Action, the Examiner has maintained his rejection of claims 57 and 60-61 under 35 U.S.C. § 102(b), as being anticipated by U. S. Patent No. 6,699,719 to Yamazaki *et al.* (hereinafter “Yamazaki”). In particular, the Examiner asserted that “Since Applicant never recites that the membranes remain in direct contact with the coating or support surface, Applicant's arguments are not found persuasive....” Applicants respectfully traverse the rejection.

As set forth in amended claim 57, the biological lipid membrane of the present invention, comprising a host lipid and a receptor of interest, is deposited into a coating of the surface of the substrate. Direct deposition of the biological lipid membrane is achieved by incubating the lipid mixture into the coated surface of the substrate array followed by drying the substrate array.

Contrary to the claimed invention, the bilayer membrane employed in Yamazaki is separated from the supporting surface by an aqueous film and, therefore, is not deposited directly to a coating of amine-presenting or silane molecules.

In view of the claim amendment and foregoing remarks, Applicants respectfully submit that Yamazaki fails as an anticipatory reference for failure to teach or suggest all of

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the recited elements of independent claim 57, as well as dependent claim 60 and 61. Thus, Applicants respectfully request that the Examiner reconsider and withdraw the rejections of these claims based upon §102(b).

Claim Rejections under 35 U.S.C. §103(a)

Claims 1, 2, 4-5, 9-16, 18, 42-50, 52, 54, 56, and 58-59 remained rejected under 35 U.S.C. §103(a) as being unpatentable for being obvious over Yamazaki and in view of Pidgeon, both as set forth hereinabove. Applicants respectfully traverse the rejection.

With respect to claims 1, 42, 52, 54, 56, and 58-59, the Examiner asserted that "Yamazaki teaches biosensor arrays comprising substrates with a plurality of distinct membranes of bilayer regions (col. 7, lines 40-50)" and that these arrays "are performed by incubating the arrays with a cholera toxin (col. 31, lines 65-67), followed by washing (col. 32, lines 1-3) and imaged with a fluorescence microscope (col. 32, lines 5-10). However, as noted by the Examiner, Yamazaki "does not teach membranes deposited on an amine-presenting molecule." To cure this deficiency, the Examiner cites Pidgeon which, according to the Examiner, teaches "a surface coated with a layer of polyamine such as polyethylenimine (PEI) for attaching membranes (col. 6, lines 32-50)" and that the functional groups such as PEI form covalent bonds with amphiphilic compounds and allow for mechanically stable structures" (Office Action at p.4, ¶2).

Applicants respectfully submit that both Yamazaki and Pidgeon fail to suggest the present invention, as set forth in amended claims 1, 42, 49, and 57 and any of the claims dependent therefrom. For example, claim 1, 42, and 49 recite that direct deposition of the biological lipid membranes is achieved by mixing a host lipid and a doped lipid together before being deposited onto the surface of the substrate and followed by air drying of the array. Similarly, in Claim 57, which was not cited by the Examiner in this rejection but is the generic claim for rejected dependent claims 58 and 59, recites that direct deposition of the biological lipid membrane is achieved by incubating the lipid mixture on to the coated surface of the substrate array and followed by drying of the array, for example by air. After deposition, the biological lipid membranes are washed, incubated under controlled humidity, and dried for 1 hour to "enable possible lateral redistribution of the lipid molecules in the supported membrane" before coming in contact with a target compound. See the specification at p. 12, ¶ [0043], l. 1-p. 13, ll. 1-11.

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As discussed above, direct deposition of a mixture of host lipid and doped lipid into the amine-presenting moiety-coated surface of the substrate array offer several benefits. First, there is no need to utilize any immobilization means or functional linkers, as disclosed by Pidgeon. Moreover, direct deposition increases the surface resistance to physical desorption and improves the membrane's long-range lateral fluidity. See the specification at pages 11-12, paragraphs 40 and 42. In fact, lateral fluidity of the membrane microarrays (slides with printed microspots and doped with fluorescently labeled lipids) according to the present invention was tested and shown using fluorescence microscopy. See the specification at p. 12, ¶ [0042].

As noted hereinabove and in various prior filed Responses, Yamazaki fails to disclose or suggest the use of biological membranes directly deposited to a coating of amine-presenting molecules, as set forth in the claims of the present invention. Yamazaki does, however, describe the fabrication of arrays of fluid bilayer membranes, but the bilayer membranes employed in Yamazaki are separated from the supporting surface by "an aqueous film of corresponding thickness" (col. 8, ll. 1-11). This aqueous film can be made of "a buffered saline solution (*e.g.*, PBS)" and can be "readily changed (taking care, of course, to keep the supported bilayer submerged at all times) by, *e.g.*, flow-through rinsing with a solution having a different composition" (col. 10, ll. 4-9). Accordingly, Applicants respectfully submit that the primary reference cited, Yamazaki, fails to disclose or suggest the direct deposition of biological membranes to a coating of amine-presenting molecules.

Applicants respectfully submit that the proposed combination with Pidgeon fails to cure the many deficiencies of Yamazaki as a primary reference. As discussed earlier, a biological membrane (*i.e.*, artificial membrane structure), as disclosed or suggested by Pidgeon, cannot be directly deposited to a coating of an amine-presenting molecule (*i.e.*, supporting material) without the use of an immobilization means. Accordingly, any biological membranes employed in Pidgeon, as with those in Yamazaki, are not directly deposited to a coating of an amine-presenting molecule and the combination fails to suggest the present invention, as claimed.

Because Yamazaki and Pidgeon neither disclose nor suggest elements of the claimed invention, *e.g.*, claims 1, 42, 49, and 57, as well as any of the claims depending therefrom, Applicants respectfully submit that the primary and secondary references do not render these claims obvious. See MPEP §2143.03 ("To establish *prima facie* obviousness of a claimed invention, all the claim limitations must be taught or suggested by the prior art").

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In addition to the individual and combined failure of Yamazaki and Pidgeon, Applicants respectfully submit that the primary reference, Yamazaki, teaches away from the invention as claimed in claims 1, 42, 49, and 52, as well as the claims dependent therefrom. In particular, Yamazaki describes that a thin polymer film (e.g., polyacrylamide or dextran) can be deposited to an array surface to form bilayer-compatible regions, and that this thin polymer film can be coupled to the array surface by 3-methacryl-oxypropyl-trimethoxy-silane (col. 18, ll. 20-29).

With respect to Pidgeon, the artificial membrane of Pidgeon is covalently immobilized or functionally linked to the substrate, which is contrary to the claimed invention. The present specification describes that “a unique aspect of the membrane microarrays is the need to keep the probe confined to the microspot while maintaining the desired lateral movement of individual molecules within the microspot—properties that are contradictory and preclude covalent immobilization of the membrane.” See the specification at page 11, ¶ [0040], line 1—page 13, lines 4-7. Based on the above, Applicants respectfully submit that Pidgeon teaches away from the claimed invention.

Based on at least the reasons presented above, Applicants respectfully submit that both Yamazaki and Pidgeon individually teach away from the claimed invention. Accordingly, one of ordinary skill in the art, [upon reading Yamazaki and Pidgeon] would be discouraged from following the path set out in this reference, or would be led in a direction divergent from the path that was taken by the Applicant.” See *In re Gurley*, 31 USPQ2d 1130 (Fed. Cir. 1994). Applicants respectfully submit that Yamazaki and Pidgeon either alone or in combination, fail to teach or suggest each and every element of the claimed invention. See MPEP 2143.03 (“To establish *prima facie* obviousness of a claimed invention, all the claim limitations must be taught or suggested by the prior art”). Accordingly, Applicants respectfully submit that claims 1, 2, 4-5, 9-16, 18, 42-50, 52, 54, 56, and 58-59, are not obvious over Yamazaki, in view of Pidgeon.

On page 5, section 21 of the Office Action, the Examiner again rejected claim 17 under 35 U.S.C. §103(a), as being unpatentable for obviousness over Yamazaki, in view of Pidgeon, as above, and in further view of U. S. Patent No. 5,004,543 to Pluskal *et al.* (hereinafter “Pluskal”). Applicants respectfully traverse the rejection.

In the Office Action, at page 6, line 2, the Examiner acknowledged that Yamazaki is deficient with respect to a microporous support. Likewise, Pidgeon fails to cure the

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deficiency. However, this deficiency, according to the Examiner, can be found in the tertiary reference. Pluskal, as asserted by the Examiner, "teaches a charge-modified, hydrophobic microporous membrane" and "further teaches that the membrane exhibits a combination of ionic and hydrophobic properties, rendering them highly effective for macromolecular adsorption applications under a variety of conditions (col. 2, ll. 35-46)." Claim 17 depends from amended claim 1.

As with the combination of Yamazaki and Pidgeon, Applicants respectfully submit that the proposed combination with Pluskal fails to cure the deficiencies of the primary and secondary references. Pluskal relates to a hydrophobic material having a crosslinked, cationic charge-modifying coating such that the majority of the ion exchange capacity of the material is provided by fixed formal positive charge groups. According to Pluskal, the pore size of the microporous membrane is at least 0.05 microns, with a maximum pore size of about 10 microns. See Pluskal at col. 3, ll. 19-20. While hydrophobic membranes can be polypropylene, polyethylene, polysulphone, PTFE or the like, PVDF membranes are preferred. See Pluskal at col. 4, ll. 64-66. The charge-modifying agent is preferably a polyamine epichlorohydrin resin. See Pluskal at col. 4, ll. 66-67. However, Pluskal does not describe any biological membrane deposited to a coating of amine-presenting molecules. Accordingly, Pluskal is outside the purview of the present invention, particularly as set forth in claim 17.

Because Yamazaki, Pidgeon, and Pluskal, either individually or in combination, fail to disclose or suggest the elements of claim 17, Applicants respectfully submit that these references do not render claim 17 obvious.

Applicants also respectfully submit that the Office Action has failed to establish any motivation to combine Yamazaki, Pidgeon, and Pluskal.

The Examiner relied upon Pluskal's teachings and concluded that "it would have been obvious to one of ordinary skill in the art to have a charge-modified, hydrophobic microporous membrane as the support in the method of Yamazaki *et al.* and Pidgeon, as suggested by Pluskal *et al.*, as the membrane is highly effective for macromolecular adsorption applications under a variety of conditions" (Office Action at p. 6, ¶2). However, Applicants again respectfully submit that, a mere statement that the combination of the prior art meets the claimed invention and would have been within the ordinary skill in the art, is not alone sufficient to establish a *prima facie* case of obviousness. See MPEP §2143.01

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(emphasis added). Accordingly, Applicants respectfully request that the Examiner provide some form of documentary proof to substantiate the alleged motivation to combine the disparate references Yamazaki, Pidgeon, and Pluskal.

Based on at least the above reasons, Applicants respectfully submit that the Office Action has failed to establish the *prima facie* obviousness of claim 17. Accordingly, Applicants respectfully request that the Examiner reconsider and withdraw the §103(a) rejection of claim 17.

Lastly, claims 51, 53, and 55 stand rejected under 35 U.S.C. §103(a), as being unpatentable over Yamazaki, in view of Pidgeon, and in further view of U. S. Patent No. 4,933,285 to Patton (hereinafter "Patton").

In the Office Action, the Examiner asserted that "Yamazaki teaches biosensor arrays comprising substrates with a plurality of distinct membranes of bilayer regions (col. 7, lines 40-50)" and that these arrays "are performed by incubating the arrays with a cholera toxin (col. 31, lines 65-67), followed by washing (col. 32, lines 1-3) and imaged with a fluorescence microscope (col. 32, lines 5-10). However, Yamazaki fails to teach a coating of γ -aminopropylsilane on the support. Likewise, Pidgeon fails to cure the deficiency. To remedy this deficiency, the Examiner states that the tertiary reference Patton teaches "substrates comprising coating of γ -aminopropylsilane (col. 4, lines 15-20)" and that "this produces solid phases that serve to anchor reaction products to a solid phase, while permitting the unreacted reagents to be removed (col. 3, lines 35-42)." Based on these statements, the Examiner concluded that Patton's teachings "would allow Yamazaki *et al.* to anchor lipid membranes to the support that have reacted with the γ -aminopropylsilane, while removing unbound lipid membranes (Office Action at page 7, lines 4-9).

Applicants respectfully submit that Yamazaki, Pidgeon, and Patton, either alone or in combination, fail to render claims 51, 53, and 55 obvious. Claims 51, 53, and 55 depend from amended claims 1, 42, and 49, respectively. The methods recited in claims 1, 42, and 49 employ a biological lipid membrane that comprises a mixture of a host lipid and a doped lipid, which are combined before being deposited onto the surface of the substrate coated with an amine-presenting molecule. After deposition, for example by sonication, the biological lipid membranes are incubated under control humidity, and dried for 1 hour before coming in contact with a target compound. None of the cited three references describes or suggests these features. Accordingly, Yamazaki, Pidgeon, and Patton, either alone or in

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combination, do not teach or suggest each and every element of claims 51, 53, and 55. Therefore, Applicants respectfully submit that Yamazaki, Pidgeon, and Patton do not render obvious claims 51, 53, and 55.

Moreover, Applicants respectfully submit that the Office Action has failed to establish a motivation to combine Yamazaki, Pidgeon, and Patton. Based on the above reasons, Applicants respectfully submit that the Examiner has failed to establish the *prima facie* obviousness of claims 51, 53, and 55. Accordingly, Applicants respectfully request that the Examiner reconsider and withdraw the §103(a) rejection of these claims.

In light of the foregoing remarks, Applicants respectfully request that the Examiner reconsider and withdraw all of the above-mentioned obviousness rejections based on §103(a).

Finally, Applicants respectfully submit that the above §§102(b) and 103(a) rejections of the pending claims have been overcome. Reconsideration and withdrawal of these rejections are earnestly requested.

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CONCLUSION

Applicants respectfully submit that this application is in condition for allowance. Favorable consideration and prompt allowance of the claims are earnestly solicited. A fee for one-month extension of time is due for filing this response. The Commissioner is hereby authorized to charge any payment deficiency to deposit account number 19-2380 referring to attorney docket number 015275-060007.

Should the Examiner believe that anything further is desired in order to place the application in even better condition for allowance, the Examiner is invited to contact Applicants' attorney of record.

Respectfully submitted,

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